WEST AFRICAN COCOA RESEARCH INSTITUTE

QUARTERLY PROGRESS REPORT

No. 42

TAFO, GOLD COAST.

PRINTED BY THE GOVERNMENT PRINTER, ACCRA.

WEST AFRICAN COCOA RESEARCH INSTITUTE
TAFO

FORTY-SECOND QUARTERLY REPORT : APRIL-JUNE, 1956

SENIOR STAFF

Director	:	J. Lamb
Secretary	:	G. H. Neal (on pre-retirement leave)
Acting Secretary	:	R. J. Webb

TAFO STATION

Division of Entomology	:	D. J. Taylor F. Raw (on leave 6th June, pending termination of secondment) P. F. Entwistle Helen M. Entwistle
Division of Pathology	:	W. T. Dale A. L. Wharton G. Usher A. Attafuah (on leave 4th June)
Division of Botany	:	A. D. McKelvie
Division of Soil Science and Chemistry	:	T. A. Rohan (on leave 18th May) E. Margaret Holden (on leave) P. W. Arnold (arrived on secondment 1st May)
Division of Agronomy	:	R. J. Benstead (on leave 23rd April)
Guide-Demonstrator	:	F. E. Decker
Station Manager	:	S. E. Ntim-Kwafo

IBADAN SUB-STATION

Acting Officer-in-Charge	:	J. Nicol (died 24th/25th June)
Entomologist	:	R. G. Donald
Pathologists	:	J. M. Thresh R. M. Lister (on leave prior to resignation).

STAFF MATTERS

It is with deep regret that we record the death of Mr. and Mrs. J. Nicol, who were amongst the victims of the Kano air disaster on 24th June, the day of their departure for the United Kingdom on leave. Mr. Nicol came to W.A.C.R.I. in April 1944, a few weeks after its inception, and was Senior Entomologist at Tafo for several years, before being transferred to the Ibadan Sub-station in 1954 as Acting Officer-in-Charge. 'Nick' and 'Bessie' will be missed by their many friends in the Gold Coast and Nigeria. Our deepest sympathy is extended to their young daughter, Sheila, and other bereaved relatives.

The Director, Mr. Lamb, flew to the United Kingdom on 3rd June and returned on July 6th. His visit was connected with recruitment of Senior Staff and other business of the Institute. Mr. W.T. Dale acted as Director, and Mr. R.G. Donald is now in charge of the Ibadan Sub-station. As should have been mentioned in the previous Quarterly Report, Mr. F.E. Decker, Senior Assistant Specialist in the Entomology Division, was appointed Guide-Demonstrator on 1st January. Mr. R.M. Lister has resigned to take up a post at the Scottish Horticultural Research Institute. We are again indebted to Rothamsted Experimental Station, this time for the secondment of Dr. P.W. Arnold (Soil Chemist) who arrived on 1st May. He will remain for approximately twelve months, and later in the year will be joined by an officer at present undergoing training in soil work. This arrangement will ensure continuity in important manurial experiments on cacao (Section 6 of this Report).

Mr. Nicol and Mr. J.M. Thresh from the Ibadan Sub-station visited Tafo in April and Mr. R.J. Webb (Acting Secretary) spent a few days at Ibadan in May. Mr. A.D. McKelvie went to Nigeria in June to arrange the planting of Variety Trials at I.N.A. and Akure.

VISITORS

Dr. S.N. Adams, former Soil Chemist, who is now with Rothamsted, spent several weeks at Tafo during May-June, to 'hand over' to Dr. Arnold. Mr. T.N. Hoblyn (see previous Quarterly Report) left at the end of May, and has submitted recommendations regarding existing and proposed field experiments. We are grateful to the West African Council for Medical Research for permitting Mr. G. Surtees to visit Tafo during June, to carry out a survey of insects of medical importance. A meeting of the Fertilizer Sub-committee of the Technical Advisory Committee for the Development of Agriculture and Natural Resources (Gold Coast) was held at the Institute in June.

Other visitors to Tafo have included Mr. J.T. Saunders (Principal of the University College, Ibadan), Mr. K. Humphreys (Secretary, West African Examinations Council), Sir William Slater (Secretary, Agricultural Research Council), Professor R.L. Wain (Wye College), Professor H.G. Baker, Dr. J.K. Morton and Mr. E.M. Nyenhuis (University College of the Gold Coast), Professor T.A. Bennet-Clark (King's College, London), Mr. C.W.S. Hartley (Acting Director) and Mr. R.A. Bull (Plant Physiologist), West African Institute for Oil Palm Research, Mr. T.S. Jones (Acting Director of Agriculture, Sierra Leone), Mr. H.D. Jordan (Officer-in-Charge, West

African .../

African Rice Research Station), Mr. J.A. Austin (Director of Agriculture, Gambia), Mr. Milam (Agricultural Attache to the American Embassy in Liberia), Mr. D.A. Chapman (Secretary to the Gold Coast Cabinet), Mr. M. Therson-Cofie (Editor, Gold Coast Daily Graphic and Sunday Mirror), and members of the Gold Coast and Nigerian Departments of Forestry, Agriculture and Agricultural Research.

Amongst visitors to the Ibadan Sub-station were His Excellency the Governor-General of the Federation of Nigeria, His Excellency the Governor of Dahomey, and Mr. Hoblyn.

MANAGEMENT AND MAINTENANCE

A Chevrolet Saloon and a Land Rover have been purchased, and the latter has been allocated to the Entomology Division.

Bungalows 15 and 16 at Tafo were re-decorated and repairs to 148 quarters at W.A.C.R.I. Village carried out. Work on the new laboratory block at Ibadan has continued throughout the quarter. All fixed wooden furnishings are now in position, and water and gas taps are being fitted.

The average daily production and consumption of potable water at the Tafo Station were as follows :-

	Production	Consumption
April ...	23,723 gallons	22,280 gallons
May ...	22,981 "	19,426 "
June ...	19,705 "	16,873 "

Samples submitted for analysis were reported to be free from coliform organisms.

CROP DELIVERY

Weekly harvesting of the Minor Crop was started on 5th June. Of the 5,197 pods harvested from the Uniformity Trial Areas, only 3 per cent. were discarded due to all causes.

SWOLLEN SHOOT DISEASE

Trees removed during treatment for swollen shoot disease are shown overleaf :-

Station .../

Station	April	May	June	Total
Old Station	63	37	71	171
Square Mile*	383	266	231	880
Adonkwanta	10	9	35	54
Akwadum	5	-	10	15
Nankese	-	-	-	-
Kukua	32	43	36	111
Koransang	20	--	41	61

* Includes suspects in addition to trees showing symptoms and their contacts.

Losses from swollen shoot disease were heavy during the quarter, only the Nankese station remaining free from it. Heavy flushing on all farms assisted diagnosis, and probably accounted for the large number of trees cut out at Kukua and Koransang.

In addition to the above, 34 infected replanted seedlings were removed from the Old Station and a further 48 trees on this area were lost from undiagnosed causes.

OUT-STATIONS

Progressive thinning of the cacao and reduction of shade where necessary have been continued. Over 60 oil-palms have been felled at Nankese. Capsid damage was slight apart from fresh attacks reported on the Resistance Trial Area at Akwadum and in Plot A at Adonkwanta. Five acres of cacao at Adonkwanta have been selected for yield recording.

METEOROLOGY

Station	Rainfall in inches				Wet days			
	April	May	June	Total	April	May	June	Total
Tafo	8.57	8.57	5.12	22.26	9	16	19	44
" 18-yr mean	5.66	6.93	8.54	21.13	11.2	13.7	18.6	43.5
Adonkwanta	3.12	6.83	9.97	19.92	4	9	8	21
Akwadum	4.18	9.84	7.94	21.96	5	6	13	24
Nankese	5.63	7.83	7.96	21.42	4	8	11	23
Kukua	3.97	7.43	7.77	19.17	5	8	9	22
Koransang	3.91	7.60	10.08	21.59	5	8	10	23

UPPER AMAZON SEED

Totals of 740 F2 Upper Amazon pods and 200 rooted cuttings have been supplied to the Western Region Department

of .../

of Agriculture, Nigeria, during the quarter. F3 pods totalling 1,830 were also sent to the Regional Departments, 1,750 in two consignments by road and 80 by air.

RESEARCH WORK

The Research Programme summarized in the Annual Report for 1954-55 is being largely followed, with certain changes in emphasis; though shortage of Senior Staff has necessitated suspension of work on some projects and hindered the initiation or expansion of others.

TAFO STATION

1. VIRUS RESEARCH

Host Range of Cacao Viruses

A few of the forms designed for obtaining information about the distribution of the baobab (Adansonia digitata L.) continue to be returned by Agricultural Survey Officers, but no further infected specimens have been located. It is well known that baobabs in the Gold Coast differ in the shape of their fruits, which may be elongate or round, but we are indebted to Professor H.G. Baker of the University College of the Gold Coast for pointing out other differences. Trees with glabrous leaves tend to come into leaf later than hairy-leafed ones, and to have narrower leaflets which are usually serrated towards the apex. These characters are poorly correlated with elongate fruits. Hybridization between the two types obviously occurs. The glabrous form can be experimentally infected with cacao and baobab viruses, but though the hairy one is probably also susceptible this requires checking.

Seedlings derived from a Ceiba devoid of spines but with rather fat pods, at Abonnu near Lake Bosumtwe, have been infected with the New Juaben and Kpeve cacao viruses. The parent tree is doubtless one of the hybrids discussed in the previous Quarterly Report (p.6). We are grateful to Professor Baker for the opinion that the tree known to us as Bombax buonopozense P. Beauv. is probably B. angulicarpum Ulbrich, and that on present evidence Pachira oleagina Decne. is a better name for the West African tree that has been identified with B. sessile (Benth.) Bakh. (Annual Report for 1954-55, p.33; Flora of West Tropical Africa, 1, Pt.2, 2nd edition). The two previously unidentified species of Cola (nos.2 and 7), are C. umbratilis and C. boxiana, both of Brennan & Keay.

Most of the host-range work during the quarter has been concentrated on the Malvaceae, but no further positive results have been obtained in this family. Attempts were made to transmit the New Juaben virus from Heliocarpus popayanensis to Triumfetta rhomboidea Jacq. and Abutilon guineense Schum. & Thonn., using the 'bottle-grafting' (modified approach-grafting) technique. Temporary union was obtained between scions of the first on stocks of the second (both are Tiliaceae), but it is not yet known whether virus has been transmitted; scions on A. guineense (Malvaceae) failed completely. After several negative tests, C. chlamydantha K. Schum. has been experimentally infected with the Bosumtwe cacao virus. Sterculia setigera Del. has been infected with the Offa Igbo virus and Theobroma angustifolia DC. with Bosomuoso (2).

Insect .../

Insect Transmission Studies

Further attempts to transmit the New Juaben virus with nymphs or young adults of Pseudococcus njalensis Laing given one hour or less on young source plants were unsuccessful. Two small-scale experiments to find out whether source feeds of more than 24 hours result in reduced infectivity, regarding which previous evidence is conflicting (Annual Report for 1954-55, p.34), gave no support to this suggestion. In the first, young adults were given access to infected plants for one, two or three days, and those found feeding at the end of the period were immediately transferred to cacao-bean test-plants at the rate of one per bean. Matters were so arranged that all the insects were transferred at approximately the same time on the same day. The respective transmission rates were 50, 55 and 43%. The second experiment, carried out concurrently with the first, was identical, except that nymphs were used. Those given one or two days' access to the source plants each infected a third of the test beans, but too few were found feeding after three days to give a valid comparison. It might be inferred from these results that young adults are slightly more efficient vectors than nymphs, but the difference is probably not a real one; in another experiment using single insects the former gave 57% transmission and the latter 54% (see also 38th and 40th Quarterly Reports).

Attempts to transmit the New Juaben virus with Dysmicoccus brevipes (Ckll.) collected from groundnuts were unsuccessful. These insects were difficult to establish on cacao. In the past, low rates of transmission of the New Juaben and Kpeve viruses have been obtained with D. brevipes from pineapples; but the specimens from groundnuts probably represent a distinct strain. Attempts to transmit the Asalu virus with P. njalensis were once again unsuccessful. Ferrisiana virgata (Ckll.) has proved to be an efficient vector of the Amafie (Western Province) virus.

Classification of Viruses and Virus Strains

It was noticed after infecting young saplings with the Asalu virus (by grafting), as a preliminary to protection tests, that leaf necrosis and abscission were usually quite severe. These seem to be early symptoms of this Nigerian virus. Observations on selected Gold Coast and Nigerian viruses have been continued. There are marked differences in virulence, which are being assessed by leaf counts, leaf product (length x breadth) measurements, and height measurements. It is a simple matter to distinguish some of the viruses but others are difficult; even when differences are noticeable they may be hard to describe concisely. Sometimes symptom sequences provide the best means of separating isolates. Several of the viruses seem to occur as complexes of strains of varying severity, as already described for New Juaben (see Posnette & Todd, Ann. appl. Biol., 43, p.433, 1955). This can make assessment of virulence rather meaningless. For example, isolates at Tafo originally derived from the Ilesha outbreak in Nigeria, though obviously variable, are in general much more severe than the immunologically related Olanla strain; but viruses from the two areas maintained at Ibadan showed quite the reverse relationship (37th Quarterly Report, p.20).

It .../

It seems likely that some cacao viruses at least have a common origin, divergence having arisen through selective adaptation in different hosts. It would be of considerable interest to demonstrate host-induced change or selection experimentally. Consequently, about eighteen months ago, what was hoped to be a relatively uniform source of the New Juaben virus was obtained in a young cacao seedling after passage through two similar plants, using single mealybugs (Pseudococcus njalensis). Virus from this source was then transferred to young plants of six susceptible species, including cacao. Since, serial passage within each host has been attempted; though, due to the resistance to infection shown by some and difficulties of seed supply in certain instances, little progress has been possible. 'Back-tests' have been made at intervals to cacao plants, which are examined for any possible host-induced variation in symptoms. So far, no evidence of change has been observed, but the experiment will be continued indefinitely.

Resistance and Tolerance

In the experiment to compare resistance and tolerance to the New Juaben virus in cuttings derived from 50 Iquitos-type F1 seedling trees (38th Quarterly Report, et seq.), the second, third and fourth replicates are at different stages towards completion. The first replicate of a similar trial to test the resistance of these trees to the Kpeve virus has been started.

Uninfested 'twinned' plants (36th Quarterly Report, et seq.) corresponding to infested ones which have either remained virus-free or shown varying degrees of tolerance to infection have now been used to provide cuttings for experiments on the lines proposed in the 38th Quarterly Report. Cuttings taken from Nanay-type plants remaining apparently uninfected after infestations made by Posnette & Todd (Ann. appl. Biol., 38, p.785, 1951) are also being rooted for comparison with Iquitos-type material as regards resistance and tolerance. Attempts are also being made to establish virus-free single-leaf cuttings from infected but tolerant plants of Nanay (loc. cit.) and Iquitos origin. The first batch of the former failed to root, perhaps because of the presence of virus. The success or failure of these attempts will depend on the extent to which the virus is systemic.

2. MEALYBUG STUDIES

A paper has recently been published by P.B. Cornwell, a former Entomologist at Tafo, entitled 'Some Aspects of Mealybug Behaviour in Relation to the Efficiency of Measures for the Control of Virus Diseases of Cacao in the Gold Coast' (Bull. ent. Res., 47, Pt.1, pp.137-166, 1956). The main conclusions of the paper are given below.

Mealybug populations (Pseudococcus njalensis) originally present in piles of slash infected with cacao viruses are usually small. It is suggested that many of the insects leave the trees at an early stage in the development of infection.

Slash piles offer conditions which induce mealybugs to remain within them. Piling retards the rate of wilting of the cut wood and consequently the rate of migration of the

mealybug .../

mealybug population. The insects continue to live and reproduce within heaps of slash for nine weeks or more. The colonies which survive longest are those in the lower part of the heaps, where death and desiccation of the material is most delayed. Reproduction is stimulated as the tissues of the felled trees become wilted, and the resulting first-instar nymphs contribute numerous participants to such migration as does occur. There is evidence from laboratory experiments that these are not potential vectors.

Mealybugs are capable of walking quite rapidly for long distances over smooth surfaces. They become incapable of movement when starved for a few days and the majority die within a week. A soil surface seriously impedes the progress of mealybugs which leave the slash heaps and this effect is more marked with the nymphal stages. Even on clean-weeded soil the rate of travel is slow and few, if any, insects can travel over appreciable distances in the period during which the virus may persist within them. Results of preliminary experiments were confirmed by the use of insects labelled with radioactive phosphorus.

Some of the ants, which become associated with slash piles and which remain in attendance on the residual mealybug population, carry these insects, but the number transported in this way is of little practical significance. The establishment of migrating mealybugs on young seedlings and mature cacao is poor, even in the presence of their attendant ants.

All these factors make the possibility of infecting healthy trees from infected slash extremely remote. There is accordingly no reason to believe that the efficiency of 'cutting out' is seriously reduced by the movement of vectors from the infected trees which are cut. Without the support of farmers for the removal of 'contact' trees, no further recommendations can be advanced to improve the present method.

3. STUDIES ON BLACK-POD DISEASE

(a) Fungicide Control Trials

Trial I is continuing for a further season but Trial II has been terminated, as it could not be expected to yield much more information. A third pilot experiment, Trial III, has been started.

The main categories of pods harvested during the quarter on Trial I are briefly summarized opposite, without giving extensive details for each treatment.

Fungicide .../

Fungicide treatment	PODS PER TREE								
	Total	Dam-aged	%*	'Black pods'	%*	Rodent-damaged	%*	Others damaged	%*
Unsprayed (Control)	5.20	4.16	80.0	0.75	14.4	2.85	54.8	0.56	10.8
Carbide-Bordeaux	3.20	2.71	84.7	0.12	3.8	2.11	65.9	0.48	15.0
Bordeaux mixture	4.44	3.96	89.2	0.47	10.6	2.89	65.1	0.60	13.5
Perenox	2.95	2.50	84.7	0.25	8.5	1.91	64.7	0.34	11.5

* Classified pods as a percentage of total harvested.

By comparison with figures for this period in 1955 (38th Quarterly Report) relatively few pods have been harvested from the sprayed plots. Carbide-Bordeaux mixture has given a similar measure of control of black-pod disease (3.8% incidence as against 14.4% for the unsprayed plots); but Bordeaux mixture (10.6%) and Perenox (8.5%) have had, as in 1954, little or no effect. If, like last year, this quarter's figures prove an indication of the control to be expected in the next two months, when the annual incidence usually reaches its peak, then carbide-Bordeaux mixture will confirm the successful results obtained with it previously but the effectiveness of Bordeaux mixture and Perenox will again be doubtful.

The control plots from Trial II are being used for Trial III, which is a simple experiment to compare two new fungicides, an oil-emulsion copper oxide - Oleocuivre (40% copper), and a new formulation of copper oxychloride - Wettable Blidust (12% copper). Perenox (50% copper) is also incorporated as a standard. Each fungicide is being used at a concentration of 0.4%, so they are being compared on an equivalent cost basis and not at equivalent concentrations of metallic copper. Only eight applications are being made, at weekly intervals from June to August.

Cushion Infection

Estimates were made last season (see previous Quarterly Report) as to how many of the proximal black-pod infections recorded on Trial I could be attributed to the fungus spreading from diseased cushions. More detailed assessment, taking pre-treatment data into consideration, has since confirmed that the lower estimates given in the Quarterly Report are more likely to be correct.

The experimental method of preventing spore infections by applying a strong fungicidal paste proved efficacious in these studies and worth repeating with slight modifications. A second experiment has therefore been started, in which trees from eight more spare control plots on Trial I (for which pre-treatment records are available) have been divided at random into two groups. One group has accessible pods painted with the fungicidal paste and inaccessible left untreated. To obtain accurate control figures for each of these categories the remaining trees are left untreated, pods harvested from them being divided into 'accessible' and 'inaccessible' and recorded separately.

Effect .../

Effect of Age and Size of Pod on Susceptibility

These new investigations are designed to study the frequency distribution of black-pod infections (proximal, lateral and distal) on pods of different ages and sizes, over a complete season, taking into account the effect of climatic factors and pod populations.

Within an area (at Tafo) where the disease is known to be severe, 200 trees are being examined every two days and on each occasion new fruits which have set are labelled and numbered. At each inspection date any fruits suffering from wilt, black-pod disease or damage due to other causes are removed, the type of injury and length and diameter of the fruit on removal being noted. Pods which reach maturity without damage are measured and recorded immediately ripening begins and then left on the trees, subject to natural disease infection, for a further period of six weeks before being harvested. If during this period they become diseased or suffer other injury they are harvested and the damage recorded.

The most note-worthy points to date are the number of cherelles attacked by the disease and the high proportion of distal and lateral infections on them. Of the 75 'black pods' harvested, 62 have been arbitrarily classified as cherelles, being under 13 cm. in length. Of these, 51 (82%) resulted from lateral or distal spore infections.

4. CAPSID STUDIES

Fog Application of BHC

Treatments have been carried out at monthly intervals in the trial which was started last quarter to find out whether capsid damage can be prevented and damaged cacao regenerated by fog application of BHC. The area consists of 10½ acres of regularly spaced mature cacao in which there are two patches of chronic capsid damage covering approximately three acres.

The insecticidal solution used in the thermal fog generator consists of 1 part 20% gamma BHC concentrate and 3 parts diesel fuel. At each treatment an average of 166 ml. of concentrate was used per acre, and the time taken to cover the whole area was about 45 minutes.

Recent capsid damage has not been found, and a considerable amount of regenerating growth is apparent on the chronically damaged trees.

Combined Cultural and Insecticidal Control

In the Costing Trial which forms part of the investigations on the Dusting and Pruning Method of capsid control (see Annual Report for 1954-55, p. 65) the frequency

of .../

of inspection and dusting has been changed from fortnightly to monthly. This change, which has been made possible by the effectiveness of the method, should considerably reduce the present cost of 6s. 8d per acre per month.

From the ~~two acres~~ under this trial 984 pods were harvested during the period of the main crop for the 1955-56 season, 68% of which were produced on the trunk and the balance on the branches.

Population Studies

Experiments to investigate the possibilities of population estimation by egg counts, which were laid down in January 1956, are continuing. A good correlation has been demonstrated between the numbers of capsids and those of unhatched eggs found in the succeeding fortnight, the importance of which is that a population estimate made by means of egg counts would give an out-of-date picture of bug numbers. A further experiment with refined technique is at present being set up at Tafo, by which it is hoped to settle the question of correlation between unhatched eggs and bugs present in the succeeding fortnight; to show, in fact, whether environmental pressure is exerted largely on post-eclosion stages rather than on eggs.

Studies on the vertical distribution of capsid eggs in mature cacao have been temporarily suspended, due to the present low population level; but they will be resumed later in the year when the effect of wider tree spacings, with more light reaching the lower parts, will be investigated.

One experiment on the rate of return of capsids to sprayed areas has been commenced and others will start in the near future.

Investigation of Fungi Associated with Capsid Lesions

Dissemination

It has been shown in laboratory experiments that spores of Fusarium decemcellulare Brick - the conidial stage of Calonectria rigidiuscula (Berk. & Br.) Sacc. - can be spread by splashing water, and by water running over sporulating tissue.

A survey of various insect species found in cacao is being undertaken to find out which are capable of carrying C. rigidiuscula. Using the plating method previously described (39th Quarterly Report), 284 insects have been examined and five shown to be contaminated. As these belong to different groups, there is unlikely to be any biological relationship between fungus and insect.

Effect of Environment on Host Reaction

After 4 weeks, watering at the rate of 500 ml. per plant per day under 50% shade increased the total number of leaves on 8-month-old cacao seedlings, compared with those given 250 ml. weekly and unshaded. Height was increased by shading after 6 weeks, and by daily watering after 7 weeks.

By .../

By this time, lesions which had developed round inoculated needle punctures were significantly longer than those around uninoculated punctures, and lesion length in daily watered plants was less than in weekly watered ones. The inhibition of lesion growth in the shaded plants was nearly significant (P=0.1).

The effect of defoliation on the growth of C. rigidiuscula and its host is being examined.

Cultural Studies

The addition of copper, zinc, iron, manganese and boron in different concentrations affects the growth of C. rigidiuscula in culture. In concentrations of 1,000 p.p.m. all these elements retarded growth; at less than 100 p.p.m. they increased the active-growth period of the fungus by 2-4 days. 1-Naphthyl-acetic acid, 3-indole-butyric acid, tryptophane, 3-indole-acetic acid, 2:4-D and copper sulphate, at varying concentrations, inhibit active growth of the mycelium; copper sulphate, tryptophane and 3-indole-acetic acid suppress growth completely at 1,000 p.p.m.

Strains of Fusarium decemcellulare

Six strains of F. decemcellulare previously isolated from cacao have been received from Dr. Harold Owen (a former W.A.C.R.I. Plant Pathologist). Their growth in culture and pathogenicity to the host are being compared.

5. EXPERIMENTS ON ESTABLISHMENT AND MAINTENANCE

Establishment Trials

The 1st Establishment Trial has been abandoned, but yield recording is continuing, as it is proposed to use the cacao for a 'Shade versus no Shade' trial. Shade removal from the proposed unshaded plots will begin at the start of the 1957 long rains. General thinning of shade over the whole area has started; alternate rows of Gliricidia have been poisoned and other dense, low-growing, indigenous trees are being removed where necessary.

On the pruned plots of the 2nd Establishment Trial chupons are now being rubbed off every fortnight, before they can become woody, instead of being cut off monthly. All shade on these plots other than Gliricidia has been poisoned. All low inter-row shade is being removed from the 3rd Establishment Trial and the cacao thinned to one plant per piquet.

Spacing Trials

Shade has been reduced in the Amelonado Spacing Trial by poisoning a large number of shade trees, including all those of Gliricidia. Chupons are now being removed fortnightly on the Upper Amazon Spacing and Pruning Trial.

Effect of Soil Type on Pod Development and Yield

Methods of Forecasting Yields of Cocoa

These investigations are continuing, but in the

absence .../

absence of an officer with sufficient statistical knowledge it has not been possible to analyse the data obtained.

Comparative Yields of Seedlings and Rooted Cuttings

This trial has been abandoned. Shortage of planting material had precluded laying out further replicates, and in the one already planted serious loss of cuttings had occurred. All the T60 material is being removed for use in a Variety Trial including certain I.C.S. clones.

Effect of Removal or Retention of Chupons on Fan Cuttings

This experiment also has been abandoned, due to the death of many plants and poor growth of the survivors. Too dense a shade may have been responsible, and it is clear that more needs to be known about the establishment of rooted cuttings under local conditions.

Frequency-of-Picking Experiments

Harvesting and recording of the 1956-57 crop have started in these experiments, which remain unaltered. Little agreement was evident between the results for 1954-55 and 1955-56; though when yields for the two seasons were combined there was an indication that, at Kukua, the 24-day harvesting treatment was slightly inferior to the others, in which harvesting is carried out at 3-, 6- and 12-day intervals.

Transplanting Experiments

The cacao in the experiment on Block P has been thinned to 8' x 8' and will now be considered as a further Routine Planting Area. The experiments in Block H have been abandoned and the land made available for other purposes.

6. SOIL FERTILITY AND MANURIAL REQUIREMENTS

Effect of Major Nutrients on Cacao

(i) Routine fertilizer applications have been made to those trials which are being continued. Measurements of seedling diameter have shown small but significant responses to superphosphate at Poanu and Juaso. A large and highly significant depression due to sulphate of ammonia has also been recorded at Poanu, though this was completely offset by superphosphate. On the Goaso trial, where growth has been particularly vigorous, the initial small response to superphosphate was no longer evident. Growth on the trials at Aiyinasi and Oyoko has been poor and there have been no responses to fertilizers. Routine applications of N, P and K have been discontinued at these two sites until more is known about the soil and environmental conditions.

On the two W.A.C.R.I. trials where shade has been removed from half the blocks, considerable damage has been caused by wood falling from dead forest trees. Further work is not considered worth while, at least until the dead trees have been felled and the final damage assessed. The response

to shade removal on these trials is still very obvious, but cannot be illustrated statistically because the experiments were originally designed as fertilizer trials without the shade-removal factor, which is inadequately replicated. Shade removal on half the blocks at Bunso initially stimulated growth but now the unshaded plants are unthrifty. The fact that grass has grown vigorously on the plots may partly account for the set-back suffered by this cacao. Shade reduction on the trials at Poanu, Juaso, Goaso and Biene is being carried out gradually.

(ii) Routine fertilizer applications have been made to the NPK trials on Upper Amazons at Asokore, Jumapo, Bunso and Anyinam. Only the Bunso trial produced a substantial yield of pods during the 1955-56 season, and there a just significant response to superphosphate has been measured. Shade is being reduced on all these trials.

(iii) The second year's pre-treatment yield recording of those areas intended for studies on the effect of fertilizers on bearing of mature cacao has been completed by the Department of Agriculture. Experiments are to be designed by 1957 for six of the areas.

The Effect of Major Nutrients from Different Sources

The 1955-56 yields from the 'types-of-N' trial at Nankese and the 'types-of-P' trial at Kabu Hill have been examined. No responses to fertilizers have been found. Shade reduction is being carried out at both sites and the experiments will be continued. Neither trial has yet produced a full crop.

Investigation of Factors Limiting Growth in the Early Life of Cacao Seedlings

Routine work has continued on this experiment.

7. VEGETATIVE PROPAGATION

The improved method of rooting cuttings under polythene sheeting (see previous Quarterly Report), which entails rooting and hardening them in the same basket, has been standardized for routine production. It is described in an Addendum to W.A.C.R.I. Technical Bulletin No.3, and a full account is being prepared for publication.

8. PLANT BREEDING

Established Trials

A number of shade trees in various trials were poisoned with 2,4,5-T during the quarter. This was effective in April but not during the heavy rains of June.

New Trials

The 3rd Clonal Trial, to test the performance of Upper Amazon selections as clones, was planted in May in Block D of the Square Mile. There are 15 selections, arranged

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so as to offset incompatibility in a randomized-block experiment with eight blocks and twelve-tree plots. The layout was designed by Messrs. Hoblyn and Pearce of East Malling Research Station. The selections in the trial are : T12.113; T16.613; T72.1767; T76.1224; T85.799; T60.885; T60.887; T60.1774; T63.882; T63.971; T79.416; T79.467; T79.501; ACU.85, and R.15.

The plants which were collected by the Anglo-Colombian Expedition in 1952-53 and introduced to W.A.C.R.I. in 1955 have been planted in a new Colombian Introductions area in Block N. Due to the few plants of each selection available, it has not been designed statistically and will be more in the nature of an Observations Area.

Variety Trials on Out-stations

Four Series II Variety Trials were planted at Oyoko, Apedwa, Pankese and Asikuma Cocoa Stations. They were planted to the same design as previous Series II trials, except at Oyoko where the layout was altered because of rocky outcrops and other obstructions. Two one-acre trials with seed at stake were planted at I.N.A. and Akure in Western Nigeria, the selections used being mainly Series I types. Pods are now being supplied for further trials in the Gold Coast and Nigeria to be planted in 1957.

Clone Collections

Five new selections have been passed as approved 'C' types. They are : C81=NA33; C82=NA34; C83=NA31; C84=IMC60; C85=PA7.

These are all parents of the approved Upper Amazon selections and as such can be planted in blocks to produce improved seed.

9. COCOA PREPARATION AND QUALITY

Effect of Fermentation on Chocolate Flavour

A number of experiments have been carried out to determine the importance of such factors as batch size, duration of fermentation and frequency of turning in the fermentation of West African Amelonado cocoa. Batch size was varied between 20 lb. and 250 lb. of wet beans; duration of fermentation was four, five or six days; frequency of turning included treatment on the second and fourth days, the third day only, and no turning. The success of the various methods was assessed by making chocolate samples which were evaluated by a local tasting panel.

Physical and chemical determinations were made during the course of each experiment, as outlined briefly below. Changes in pH of the cotyledons and pulp were followed, and time-temperature curves plotted from readings given by thermometers placed at the top, centre and bottom of each fermenting mass. Variations in moisture content of the beans were determined, and the polyphenolic compounds estimated daily by quantitative chromatographic analyses on paper. In the following summary of results no experimental details are given, as these will

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be published elsewhere.

It has been demonstrated that as little as 20 lb. of wet cocoa can be successfully fermented in a period of four days without mixing the heap. Larger batches require longer fermentation and mixing is essential for uniformity. In very large batches, of 600 lb. and over, it is observed on opening the fermenting mass after 48 hours that there is an outer layer of beans which exhibit changes in appearance associated with fermentation, whereas the central core is quite unaltered. Presumably, those beans at the centre will, after mixing, start to undergo changes and in six days will have had the equivalent of four to five days fermentation. Air penetration of the fermenting mass is necessary to promote bacterial oxidation of the alcohol formed by fermentation of the pulp sugars. With small batches such penetration is more complete, and the beans will all die at about the same time as a result of increased temperature and acidity; in larger batches the central beans may not die until 48 hours after those at the surface. This probably accounts for the lack of uniformity in appearance of beans fermented in large quantities.

Quantitative chromatographic analyses of the polyphenolic compounds during fermentation have shown that whereas the anthocyanins are removed during fermentation the catechins and leucocyanidins are not greatly affected until the drying stage, when they are attacked by an oxidase system. It is interesting to note that the removal of catechins and leucocyanidins on drying is independent of their concentration after fermentation; and that the anthocyanin concentration is reduced during the initial stages of drying, suggesting a continuation of the fermentation process while the moisture content of the beans is still high.

Temperature measurements indicate that the opinion of some authorities, who hold that the fermenting mass must achieve a minimum temperature of 50°C., may not be strictly valid. During the present investigation a satisfactory product resulted in a number of experiments where the temperature did not reach this recommended value.

Measurements of pH during fermentation and drying have shown a reduction for the cotyledons (6.5 to 5.1) and an increase for the pulp (3.6 to 6.0). On drying, there was a slight pH increase in the cotyledons. Those beans which had four or five days' fermentation had a cotyledon pH of 5.4 after drying, whereas after six days the value was very slightly higher (5.7). Such small differences have no significant effect on flavour.

During fermentation there was quite a distinct increase in moisture content of the beans, from an initial 34.0% to 39.5% after six days. Since this increase must arise through absorption from the pulp it is highly probable that the pulp has an effect on flavour.

The external appearance of the beans after fermentation was carefully noted. It was observed that those fermented for only four days were clean and reddish brown in colour, while those fermented for six were dirty in appearance and dark brown. Internally, the beans from four-day fermentations tended to be more purple, but they gave good chocolate

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on processing. It is worth emphasizing that these beans, though classed as purple, contained only a very small fraction of the anthocyanins initially present, as proved by quantitative chromatographic analyses.

Drying of Wet Fermented Cocoa

An introductory investigation was made into this field during the last crop season. In general, it was found that the beans were sufficiently dry for bagging after seven to nine days. The reduction in moisture content on further drying is of little advantage as the beans re-absorb moisture on storage.

Quantitative analyses of the polyphenols during drying have shown that the greatest changes take place during the first week, there being no significant drop in their concentration during the second and third weeks. Additional information on the behaviour of these compounds during drying has been given above.

Influence of Storage on Purple Beans

Results obtained from experiments in which cocoa has been stored under different atmospheric conditions show, so far, that the only noticeable effect is on moisture content. A slight drop in purple-bean count has admittedly been observed in a number of instances but this could be due merely to sampling error. In any event, it is now fairly certain that even if a reduction in purple beans is observed it represents only a very small change in the actual percentage of anthocyanins present, and may well have no significant effect on flavour.

A Survey of Farmers' Fermentation Methods

Samples and questionnaires have been received from only 214 of the 300 selected farms. The samples have been graded and the data await the appointment of a statistician, although it is possible that the poor response has invalidated the investigation.

11. INSECTICIDE AND FUNGICIDE STUDIES

Pollination Studies

In the previous Quarterly Report this work was erroneously considered under Plant Breeding (8). Although information of value to Cacao Breeders is being obtained, the ultimate aim is to estimate the effect of insecticidal spraying on pollination and yield.

(i). Entomological. From routine collections of insects in cacao flowers a fair number of Ceratopogonids have been obtained, comprising three species, of which one is very much more common than the other two. Cecidomyids are still being found in about 3% of flowers, and recently small but regular numbers of Drosopholids, Collembola and parasitic Hymenoptera have been recorded.

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A large number of samples of decaying vegetable matter of all kinds, from the Square Mile, have been kept in the insectary, and from them a number of Ceratopogonids of the same genera as those from cacao flowers have been obtained; but these have not yet been identified as to species.

The period of the day during which most pollination takes place was investigated during April and May, a total of 5,779 flowers being examined. The daylight hours were divided into 5 periods, and it was established that there are two peaks of pollination, of which that in the morning is much the larger :-

Period of the day	0600-0730	0730-0900	0900-1200	1200-1500	1500-1800
Percentage of total pollination	37.1	49.5	1.0	12.4	0

Heavy rainfall in the early morning means that very little pollination will take place that day, while afternoon storms hardly affect it.

(ii). Botanical. Hand pollinations to determine whether fertilization and setting can result from pollen deposited at the base of the style were continued on several trees in the Old Station. A powerful hand lens was used to examine the stigma and style and ensure that the pollen was in the correct place. The results show that fertilization can take place as easily through the style as through the stigma :-

	No. of flowers	No. set	% Setting
Pollinated on stigma	28	21	75.0
Pollinated on style	31	27	87.1

A further series of hand pollinations is being carried out to compare the proportion of pollinated flowers which set on branches of differing diameter.

A long-term experiment, using 36 trees of the guard-row of the 2nd Clonal Trial, was started in May to compare pollination and setting rates throughout the year.

12. PHYSIOLOGY OF NUTRITION, GROWTH AND FRUITING

The experiments described in the previous Quarterly Report are continuing but few results are yet available.

Investigation .../

Investigation of Cherelle Wilt

The Effect of Increased Setting on Yield

Increased setting is being induced by large numbers of hand pollinations, 778 of which have already set on the twenty trees of the experiment. Figures for the period February - June (inclusive) are as follows :-

	Hand-pollinated trees (20)	Natural-pollinated trees (20)
Pods set	2049	1536
Pods wilted	1367	761
Pods harvested	31	78
% wilted	66.7	49.5
Pods on trees at 30/6/56	904	704

Increasing the number of sets on the trees has increased the percentage of wilt and reduced the number of pods harvested. It is too early to estimate the effect on final yield.

Loss of Nutrients from Wilting Pods

The following figures illustrate the changes in weight of a wilting cherelle of about 6 cm. long, from the time decrease in growth is first apparent until five weeks later :-

Date	Symptom	% Water content	Dry weight (g.)	Weight of ash (g.)
2/4/56	Green healthy cherelle	86.2	2.0	0.19
9/4/56	Yellowing appears	86.1	1.9	0.19
16/4/56	Turns brown	82.4	1.4	0.15
23/4/56	Turns black and shrivels	52.3	1.1	0.11
30/4/56	Becomes hard	47.1	1.2	0.11
7/5/56	" "	36.8	1.2	0.11

Cherelles lose dry matter and ash in the first two to three weeks after wilting and this probably means that some nutrients are withdrawn into the tree. At present wilting cherelles are being analysed for N, P and K.

Experiments .../

Experiments with Radioactive Phosphorus

Work is now in progress, using P^{32}, to study the absorption and translocation of minerals in cacao trees. Trunk injection has given rapid uptake in young seedlings under conditions producing a high transpiration rate; in those under shade or in humid conditions, and in mature trees in the field, there was no uptake with either trunk injection or soil application. During these experiments both soil and atmosphere had low saturation deficits and transpiration was at a minimum. Before any more work is done on the utilization of P^{32} in the tree, an experiment will be carried out to study the rate of uptake from the soil throughout the year, to find the best time for soil applications.

It has been shown that pods with their stalks immersed in liquid rapidly take up P^{32}, but that there is no absorption when it is applied externally to the pod or to the stalk, either in lanolin or aqueous solution. This may explain the lack of response of pods to applications of growth substances.

IBADAN SUB-STATION

1. VIRUS RESEARCH

Virus Strains

A number of Nigerian isolates of cacao virus are being studied intensively. The work includes protection tests, experiments to investigate the effect of the various isolates on growth, and detailed observations on symptom production. Important differences are being found and it appears that the range of viruses in Nigeria is greater than originally thought. Field observations continue and yield records are now being taken on three plots to investigate the effect of virus infection on yield.

Coppicing Experiments

Routine observations have continued on the various coppicing experiments at the I.N.A. Farm near Ibadan, and at Owena in Ondo Province. No further coppicing experiments will be started and the detailed aspects of this work have already been completed.

The results of the I.N.A. experiment, which was designed to investigate seasonal effects, have recently been analysed in detail. It has been found that the rapidity with which trees regenerate after coppicing is correlated with the weather conditions in the period following this treatment. They regenerate most rapidly after coppicing in the dry season, and under these conditions the first chupons are to be found on the stumps about four weeks later. The rate of chupon production falls with increase in rainfall and decrease in temperature in the weeks after coppicing, and in the cool humid period at the height of the rains chupons are not found

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on the stumps until after about 7 weeks. Multiple correlation analyses suggest that temperature and rainfall are each having significant and independent effects on the rate of chupon production. This result is consistent with observations made by other workers, who found that conditions were relatively unfavourable for the growth of mature cacao at the height of the rains (Humphries, Ann. Bot. (N.S.), 8, p.259, 1944; Greenwood & Posnette, J. hort. Sci., 25, p.164, 1950).

4. CAPSID STUDIES

Sixty-three plots suitable for the proposed insecticidal control trials have been selected, their boundaries marked and 100 trees in each labelled for detailed recording. These plots are arranged in nine groups of seven, three groups being near Araromi in the south of Ibadan Province, three between Ife and Ilesha in Oyo Province, and three between Owena and Aponmu in Ondo Province. Within each group, the following seven treatments have been allocated at random to the plots :-

Treatment	Dosage of gamma BHC	Method	Frequency of application
A	2 oz./acre	Low vol.	Every 4 weeks from Aug. to Feb. inclusive
B	4 oz./acre	Low vol.	Every 4 weeks from Aug. to Feb. inclusive
C	4 oz./acre	Low vol.	Every 8 weeks from Aug. to Feb. inclusive
D	4 oz./acre	High vol.	Every 4 weeks from Aug. to Feb. inclusive
E	4 oz./acre	Low vol.	Aug. and Sept., followed by high-volume monthly applications where damage occurs
F	4 oz./acre	Hand duster	Aug. and Sept., followed by monthly hand-dusting applications where damage occurs.
G	NO TREATMENT		

All plots are to be sprayed with Perenox against black-pod disease. This was begun in Ondo and Oyo Provinces on 21st May and in Ibadan Province on 18th June, and it will be continued until the end of the harvest. Recording was started on 18th June in all plots.

MINOR PESTS OF CACAO

Attacks by larvae of a Limacodid moth were reported from several localities in the Ibadan Province during May. The larvae were found to be stripping the leaves from cacao,

oil-palms .../

oil-palms, bananas and other plants, but were found to be more or less confined to relatively small isolated areas, in some of which severe defoliation occurred. Some of the older farmers attributed these attacks to the prolonged dry season which has been experienced this year and stated that the pest would disappear with the onset of the rains. This seems to have occurred.

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